

CLAIMS

1/ DNAs, characterized in that they are in all or part genes, with their reading frame, present in *Neisseria meningitidis* (called Nm below), but absent both from *Neisseria gonorrhoeae* (called Ng below) and from *Neisseria Pectamica* [sic] (called Nl below), with the exception of genes involved in the biosynthesis of the polysaccharide capsule, *frpA*, *frpC*, *opc*, *porA*, rotamase, the sequence IC1106 [sic], IgA proteases, pilin, pilC, proteins which bind transferrin and opacity proteins.

2/ DNAs according to claim 1, characterized in that they are present in Nm, but absent from Ng.

3/ DNAs according to claim 2, characterized in that they comprise one or more sequence(s) present on the chromosome of Nm Z2491 between *tufA* and *pilT*, or region 1 of the chromosome, and/or the nucleotide sequence(s) capable of hybridizing with the said sequence(s).

4/ DNAs according to claim 2, characterized in that they comprise one or more sequence(s) present on the chromosome of Nm Z2491 between *pilQ* and λ 740, or region 2 of the chromosome, and/or the nucleotide sequence(s) capable of hybridizing with the said sequence(s).

5/ DNAs according to claim 2, characterized in that they comprise one or more sequence(s) present on the chromosome of Nm Z2491 between *argF* and *opaB*, or region 3 of the chromosome, and/or the nucleotide sequence(s) capable of hybridizing with the said sequence(s).

6/ DNAs according to claim 3, characterized in that their sequence corresponds in all or part to SEQ ID No. 9, 13, 22 or 30, and/or to any sequence located at more or less 20 kb from these SEQ ID on the chromosome of an Nm strain, and/or is

capable of hybridizing with at least a fragment of any one of these sequences.

7/ DNAs according to claim 4, characterized in that their sequence corresponds in all or part to SEQ ID No. 1, 2, 4, 6, 7, 10, 15, 31 or 34, and/or to any sequence located at more or less 20 kb from these SEQ ID on the chromosome of an Nm strain, and/or is capable of hybridizing with at least a fragment of any one of these sequences.

8/ DNAs according to claim 4, characterized in that they are all or part of the DNA sequence SEQ ID No. 36 or sequences corresponding to the open reading frames SEQ ID No. 37, SEQ ID No. 38, SEQ ID No. 39, SEQ ID No. 40, SEQ ID No. 41, SEQ ID No. 42, SEQ ID No. 43, SEQ ID No. 44, SEQ ID No. 45 and/or to any sequence located at more or less 20 kb from these SEQ ID on the chromosome of an Nm strain, and/or is [sic] capable of hybridizing with at least a fragment of any one of these sequences.

9/ DNAs according to claim 5, characterized in that their sequence corresponds in all or part to SEQ ID No. 8, 21, 23, 25, 26, 28, 29, 32 or 35, and/or to any sequence located at more or less 20 kb from these SEQ ID on the chromosome of an Nm strain, and/or is capable of hybridizing with at least a fragment of any one of these sequences.

10/ DNAs according to claim 2, characterized in that their sequence corresponds in all or part to SEQ ID No. 3, 5, 11, 12, 14, 16, 18, 19, 20, 24, 27 or 33, and/or to any sequence located at more or less 20 kb from these SEQ ID on the chromosome of an Nm strain, and/or is capable of hybridizing with at least a fragment of any one of these sequences.

11/ DNAs according to claim 1, characterized in that they are common with those of Ng, but are absent from N1.

12/ DNAs according to claim 11, characterized in that they comprise one or more sequence(s) present on the chromosome of Nm Z2491 between arg J and reg F, or region 4 of the chromosome, and/or the nucleotide sequence(s) capable of hybridizing with the said sequence(s).

13/ DNAs according to claim 11, characterized in that they comprise one or more sequence(s) present on the chromosome of Nm Z2491 between the marker lambda 375 to pen A, or region 5 of the chromosome, and/or the nucleotide sequence(s) capable of hybridizing with the said sequence(s).

14/ DNA according to any one of the preceding claims, characterized in that it codes for a protein exported beyond the cytoplasmic membrane.

15/ DNAs according to any only of claims 1 to 14, characterized in that all or part of their sequence corresponds to a region conserved within the Nm species.

16/ DNA according to any one of claims 1 to 15, characterized in that it is inserted in a transfer or expression vector, such as a cosmid, plasmid or bacteriophage.

17/ Host cell, more particularly bacterial cell or Nm cell, transformed by insertion of at least one DNA according to any one of claims 1 to 15.

18/ Cell comprising genes or gene fragments specific to Nm, more particularly bacterial cell or Nm cell, the chromosome of which is deleted by at least one DNA according to any one of claims 1 to 15, in particular a DNA responsible for the pathogenicity.

19/ DNAs, characterized in that their sequence corresponds in all or part to the transcription of at least one DNA sequence or sequence fragment according to any one of claims 1 to 15.

20/ Antisense nucleic acids, characterized in that their

sequence corresponds to the antisense of at least one nucleotide sequence according to any one of claims 1 to 15 or 19, or a fragment of such a sequence, and in that they carry, where appropriate, at least one chemical substituent, such as a methyl group and/or a glycosyl group.

21/ Polypeptides, characterized in that they have an amino acid chain corresponding to all or part of a sequence coded by the nucleic acids defined in any one of claims 1 to 15 or 19, or deduced from sequences of these nucleic acids, with, where appropriate, modifications with respect to the coded or deduced sequences, where these modifications do not alter the biochemical properties observed in the natural polypeptide.

22/ Peptides according to claim 21, characterized in that they are peptides exported beyond the cytoplasmic membrane, more specifically peptides corresponding to all or part of those coded by a DNA according to claim 14.

23/ Antibodies, characterized in that they are polyclonal or monoclonal antibodies directed against at least one epitope of a peptide according to claim 20 or 21, or fragments of these antibodies, more particularly fragments Fv, Fab, Fab'2, or also anti-antibodies capable of recognizing, by a reaction of the antigen-antibody type, the said antibodies or their fragments.

24/ Process for obtaining *Neisseria meningitidis*-specific DNA banks, comprising

- mixing of two DNA populations,
- realization of at least one subtractive hybridization-amplification iteration, and
- collection of the desired DNA or DNAs, followed, where appropriate, by its/their purification with elimination of redundant sequences.

25/ Process according to claim 24, characterized in that, to obtain a bank which is specific to Nm, in contrast to Ng

- two DNA populations originating respectively from a strain of *Neisseria meningitidis*, or a reference strain, for which the specific bank is to be constructed, and a strain of *Neisseria gonorrhoeae*, or a subtraction strain, the DNA sequences of these strains being those obtained by

. - random shearing of the chromosomal DNA of the subtraction strain, in particular by repeated passage through a syringe, and

. cleavage of the chromosomal DNA of the reference strain, preferably by a restriction enzyme producing fragments less than about 1 kb in size, and in that to obtain a bank of DNAs common between Nm and Ng, but specific with respect to Nl, three different banks are constructed, two of them by digestion of the chromosomal DNA of Nm by *MboI* and *Tsp5091*, and the third by digestion of the chromosomal DNA of Nm with *MspI*, two subtraction series are carried out, and the DNAs having the required specificity are collected.

26/ Banks of DNA clones obtained by carrying out the process according to claim 24 or 25.

27/ Use of the process according to claim 24 to obtain banks of DNAs specific to a given cell or to a given variant of the same species of cell, where another species or another variant which is close genomically and expresses different pathogenic potencies exists, in particular banks of DNAs specific to cryptococci, *Haemophilus*, pneumococci or also *Escherichia*.

28/ Method for diagnosis of a meningococcal infection, and more particularly of meningococcal meningitis, by demonstration of the presence of *Neisseria meningitis* in a biological sample, characterized in that it comprises the

stages of:

- bringing into contact a biological sample to be analysed and a reagent formulated from at least one nucleic acid as defined in one of claims 1 to 15 or 19, if appropriate in the form of a nucleotide probe or a primer, or, as a variant, from at least one antibody or a fragment of an antibody, as defined in claim 23, under conditions which allow respectively hybridization or a reaction of the antigen-antibody type, and

- detection of any reaction product formed.

29/ Method for diagnosis of an immune reaction specific to meningococcal infection, characterized in that it comprises the stages of:

- bringing into contact a biological sample to be analysed and at least one polypeptide according to any one of claims 21 or 22 or an anti-antibody according to claim 23, or a fragment thereof, these products being labelled, where appropriate, under conditions which allow a reaction of the antigen-antibody type to be effected, and

- detection of any reaction product formed.

30/ Kits for carrying out a method according to any one of claims 28 or 29, characterized in that they comprise

- at least one reagent as defined in claim 28 or 29, that is to say of the nucleic acid, antibody or peptide type,

- products, in particular markers or buffers, which enable the intended nucleotide hybridization reaction or immunological reaction to be carried out, as well as use instructions.

31/ Vaccine composition including in its spectrum, in particular in combination with at least one childhood vaccine, antimeningococcal prophylaxis and intended for prevention of any form of infection by *Neisseria meningitidis*, characterized

in that it comprises, in combination with (a) physiologically acceptable vehicle(s), an effective amount:

- of peptide according to claim 21 or 22, or
- of antibody or anti-antibody fragment according to claim 23,

this material optionally being conjugated, in order to reinforce its immunogenicity, with a carrier molecule such as a poliovirus protein, tetanus toxin, protein produced by the hypervariable region of a pilin.

32/ Vaccine composition including in its spectrum, in particular in combination with at least one childhood vaccine, antimeningococcal prophylaxis and intended for prevention of any form of infection by *Neisseria meningitidis*, characterized in that it comprises, in combination with (a) physiologically acceptable vehicle(s), an effective amount:

- of nucleic acids according to any one of claims 1 to 15 or 19 or
- of cells according to claim 17 or 18.

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